(Amended) 4. A method for measuring the amount of whole parathyroid hormone in a sample comprising:

a) adding to the sample a first labeled antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), for whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in this sequence are part of an antibody reactive portion of the peptide, in an amount sufficient to bind all whole parathyroid hormone present;

b) allowing the labeled antibody to bind to any whole parathyroid hormone present, thereby forming a complex; and

c) measuring the amount of labeled complex, and thereby determining the amount of whole parathyroid hormone.

(Amended) 5. The method of Claim 4 wherein the labeled anti-parathyroid hormone antibody or antibody fragment is a monoclonal antibody.

(Amended) 6. The method of Claim 4 wherein the labeled anti-parathyroid hormone antibody or antibody fragment is a polyclonal antibody.

(Amended) 7. The method of Claim 4 wherein a second antibody is added which is bound to a solid support and specifically binds to a portion of whole parathyroid hormone other than the initial peptide sequence which binds to the first antibody.

(Amended) 10. The method of Claim 4 wherein the label or signal generating component of the first antibody is selected from the group consisting of chemiluminescent agents, colorimetric agents, energy transfer agents, enzymes, fluorescent agents, and radioisotopes.

(Amended) 11. The method of Claim 7 also comprising adding to the sample a third antibody specific for any parathyroid hormone C-terminal fragment present in the sample, but which is not

reactive to the initial peptide sequence, thereby reducing binding reaction interference from any parathyroid hormone C-terminal fragments present in the sample.

(Amended) 12. A method for measuring the amount of whole parathyroid hormone in a sample comprising:

a) adding to the sample a first antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), for whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in this sequence are part of an antibody reactive portion of the peptide, in an amount sufficient to bind all whole parathyroid hormone present;

B3

b) allowing the first antibody to bind to any whole parathyroid hormone present, thereby forming a complex;

c) labeling the complex by means of adding a second antibody that has a label or signal generating component attached thereto and that specifically binds to a portion of whole parathyroid hormone other than the initial peptide sequence which binds to the first antibody; and

d) measuring the amount of labeled complex, and thereby determining the amount of whole parathyroid hormone.

(Amended) 15. The method of Claim 12 wherein the second labeled antibody binds either to [the] a mid-portion of whole parathyroid hormone or the C-terminal of whole parathyroid hormone and also comprising adding at least a third antibody which specifically binds to the first antibody, thereby forming a precipitating mass.

B4

(Amended) 16. The method of Claim 15 wherein the third antibody is bound to a solid support.

(Amended) 17. A method for measuring whole parathyroid hormone by means of a precipitating or turbidometric immunoassay comprising:

a) adding to a sample an antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), for whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in this sequence are part of an antibody reactive portion of the peptide, in an amount sufficient to bind all whole parathyroid hormone present, said antibody being attached to a colloidal particle or moeity which can be used to detect a signal change;

b) allowing the antibody to bind to any whole parathyroid hormone present, thereby forming a complex; and

B4

c) measuring the change in signal due to the formation of the complex, and thereby determining the amount of whole parathyroid hormone.

(Amended) 18. A kit containing reagents for performing an assay for whole parathyroid hormone comprising:

a) a first substantially pure antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), of whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in this sequence are part of an antibody reactive portion of the peptide; and

b) a labeling component that binds to whole parathyroid hormone, but not to the initial parathyroid hormone peptide sequence.

(Amended) 19. The kit of Claim 18 also comprising a second antibody specific for any parathyroid hormone C-terminal fragment present in the sample.

(Amended) 20. A kit containing reagents for performing an assay for whole parathyroid hormone comprising:

a) a first substantially pure antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in

this sequence are part of an antibody reactive portion of the peptide having a signal generating component attached thereto; and

b) a second antibody that binds to whole parathyroid hormone, but not to the initial parathyroid hormone peptide sequence which is bound to a solid support.

(Amended) 21. The kit of Claim 20 also comprising a third antibody that specifically binds to an epitope left open after whole parathyroid hormone binds to the first antibody and the second antibody, thereby forming a precipitating mass.

B4

(Amended) 22. A method for measuring the amount of functional N-terminal parathyroid hormone fragment and whole parathyroid hormone in a sample comprising:

a) adding to the sample a first antibody or antibody fragment specific for an initial peptide sequence of amino acids 1 to 7, (SEQ. ID NO. 1), for whole parathyroid hormone which comprises a domain for adenylate cyclase activation, wherein at least four amino acids in this sequence are part of an antibody reactive portion of the peptide, in an amount sufficient to bind all functional N-terminal parathyroid hormone fragment and whole parathyroid hormone present;

b) adding to the sample a second antibody or antibody fragment specific for a peptide sequence of amino acids 28 to 34, (SEQ ID NO. 2), which comprises a domain for protein kinase C activation, wherein at least four amino acids in this sequence are part of the antibody reactive portion of the peptide, in an amount sufficient to bind all functional N-terminal parathyroid hormone fragment and whole parathyroid hormone present, at least the first antibody or the second antibody is labeled;

c) allowing the first antibody and second antibody to bind to any N-terminal parathyroid hormone fragment or whole parathyroid hormone present, thereby forming a complex; and

d) measuring the amount of labeled complex, and thereby determining the amount of whole parathyroid hormone.

(Amended) 23. A method for differentiating between a person having substantially normal parathyroid hormone function and having hyperparathyroidism comprising measuring whole parathyroid hormone.

B4

(Amended) 24. A method for differentiating between a chronic uremia patient having substantially normal active parathyroid hormone levels and having hyperparathyroidism comprising measuring whole parathyroid hormone.

Indefiniteness Rejection

Claims 1-24 have been rejected under 35 USC 112, second paragraph as being indefinite for failing to point out and distinctly claim the subject matter which applicant regards as the invention. Portions of Claims 1-24 are said to lack antecedent basis.

Response

Claims 1, 4 to 7, 10 to 12, and 15 to 24 have been amended to provide an antecedent basis for all of the claimed elements as pointed out by the Examiner.

Lack of Novelty Rejection

Claims 23 and 24 have been rejected under 3 USC 102(b) as being anticipated by a 1996 article by Brossard *et alia*. The Examiner states that Brossard determined levels of total parathyroid hormone (PTH) by two-site immunoassay and also determined the proportions of intact PTH (1-84) and a non-(1-84)PTH fragment. That study has been conducted for normal

patients and for those having renal failure (with and without secondary hyperparathyroidism.

Response

Claims 23 and 24 are novel. These claims cover differentiating normal patients from those with hyperparathyroidism by measuring not intact PTH (I-PTH), but whole PTH, that is, PTH 1-84 without other large PTH fragments.

Brossard discloses that I-PTH assays measure two peaks, PTH 1-84 and a smaller "non-(1-84)" PTH molecular form. The differentiation of these two peaks is done by HPLC fractionation. In studying the occurrence of non-(1-84) PTH between normal patient and those with renal failure, Brossard notes that this secondary peak accounts for about 21% of the I-PTH result in hypercalcemic patients and about 10% in hypocalcemic patients. As opposed to normal patients, renal failure patients had a 2.5 fold increase in the presence of this secondary peak.

Brossard does not anticipate the present claims because it does not differentiate normal patients from those having hyperparathyroidism by measuring PTH 1-84. Instead, Brossard differentiates by indirectly measuring non-(1-84) PTH. The secondary peak is the differentiating peak, not the primary peak. Nowhere does Brossard disclose simply using PTH 1-84 to differentiate between the two claimed populations.

First Obviousness Rejection

Claims 1 to 4, 6 to 21, and 23 to 24 have been rejected under 35 USC 103(a) as being obvious in view of an article by LePage *et alia*. In particular, the Examiner has stated that LePage uses two-site I-PTH immunoassays, in combination with HPLC fractionation, to determine I-PTH and non-(1-84) PTH. Allegedly, LePage characterized the non-(1-84) PTH as

similar, if not, identical to commercially available 7-84 PTH fragment. Furthermore, LePage is said to suggest that the non-(1-84) fragment lacks adenylate cyclase activity. The Examiner concludes that the claims as a whole are *prima facie* obvious absent evidence to the contrary.

Response

Claims 1 to 4, 6 to 21, and 23 to 24 are not obvious in view of LePage.

The present invention has unexpected benefits and utilities that are neither suggested nor recognized by the cited art. Support for the following statements can be found in the attached Declaration by Dr. Ping Gao.

The present invention uncovers an unexpectedly high level of non-whole PTH in normal patients. On average, up to about 50% of an I-PTH value in normal patients can be attributed to non-whole PTH. This unexpectedly high level has direct consequences in that some normal patients are interpreted by I-PTH to be on the threshold of hyperparathyroidism (and thereby subjected to increase monitoring and the associated costs), but actually the patients have normal functioning parathyroid systems that do not need such monitoring. Nowhere does LePage suggest that I-PTH values can be elevated to such a degree in normal patients.

The present invention unexpectedly is not in direct correlation with the I-PTH assays. In other words, the whole PTH assay is not simply a consistent percentage value (from less than 10% to over 90%) of the I-PTH value. Nowhere does LePage suggest that not detecting non-(1-84) PTH would lead to such variability from patient to patient.

The present invention also allows the nephrologist to avoid unwittingly overdosing secondary hyperparathyroidism patients by using standard of care PTH suppressive therapy. In the past, I-PTH assays have been unable to assess low bone turnover disease from high bone

turnover disease if the patient is in the gray diagnostic zone of 100 to 400 pg/ml. Inaccurate assessment leads to unnecessary therapy. Certain patients having an apparently high PTH value are given PTH suppressive therapy, where in reality the patient has a normal 1-84 PTH value (whole PTH). Thus, the patient is induced into an adynamic low bone turnover, tending to lead to renal osteodystrophy and soft tissue calcification, in particular coronary calcification.

A fourth unexpected benefit of the present invention is the ability to detect almost all (96%) of the primary hyperparathyroidism patients in the mild (or early onset stage) as opposed to the I-PTH ability to detect only 72% of such patients. (One should note that this magnitude of change in discrimination is equivalent to that which occurred in a recognized major clinical advance between the earlier mid-PTH assays and I-PTH assays.) This greater ability is especially important in view of the need to detect such patients as soon as possible so as to avoid irreversible loss of bone mass and organ (renal and cardiac) damage due to a high calcium blood level. Nowhere does LePage suggest that the non-(1-84) PTH fragment masks the ability to discriminate amongst such patients. Nowhere does LePage suggest that such an enhanced degree of discrimination is possible.

A fifth unexpected benefit of the present invention is the ability for the clinician to avoid suspecting malignancy associated hypercalcemia (MAH), cancer, in patients that, in reality, have primary hyperpararthyroidism. For 24% of those patients with elevated blood calcium, I-PTH assays have led to suspecting MAH and resulted in a regimen of unnecessary cancer diagnostics and a delay, if not outright denial, in providing surgical therapy. Nowhere does LePage suggest that the non-(1-84) PTH fragment plays any role in such differentiation and provide a means to avoid improper treatment.

A sixth unexpected benefit of the present invention is increased timeliness in assessing the success of parathyroidectomy surgeries. The present invention is a faster intraoperative marker, being over five minutes faster than I-PTH assays in providing the waiting surgeon with an answer

as to whether or not the excision is complete. Nowhere does LePage suggest that an assay detecting whole PTH would be a faster intraoperative marker.

Second Obviousness Rejection

Claims 1 to 21 and 23 to 24 have been rejected under 35 USC 103(a) as being obvious in view of an article by LePage *et alia* in view of a reference by Campbell. The Examiner reiterates the statements in the first obviousness rejection and also states that while LePage does not provide for monoclonal antibodies, Campbell references the general procedure for producing monoclonal antibodies.

The Examiner states that it would have been obvious to elicit monoclonal antibodies to amino acid residues 1-6 for PTH. The art is said to establish a *prima facie* case of obviousness, absent evidence to the contrary.

Response

Claims 1 to 21 and 23 to 24 are not obvious in view of Campbell and LePage. Applicants reiterate all of their arguments in the Response to the first obviousness rejection. Campbell does not suggest any of the benefits articulated in the Response.

Third Obviousness Rejection

Claims 1 to 5, 7 to 10, 12 to 14, 17, 18, 20, and 22 have been rejected under 35 USC 103(a) as being obvious in view of an article by Gao *et alia* in view of LePage. In particular, the Examiner has stated that Gao teaches a two-site immunoassay using monoclonal antibodies to

detect I-PTH and large biologically active N-terminal fragments of PTH. The Examiner concludes that it would have been obvious for one of ordinary skill in the art to elicit 1-6 antibodies in view of Gao and to use them in an assay allegedly suggested by LePage.

Response

Claims 1 to 5, 7 to 10, 12 to 14, 17, 18, 20, and 22 are not obvious in view Gao and LePage.

Gao is concerned with detecting the presence of N-terminal PTH fragments, namely, PTH 1-34 to PTH 1-38. Gao uses a two-site immunoassay to detect such fragments, employing two (not just one) anti PTH 1-38 monoclonal antibodies.

Applicants reiterate all of their arguments in the Response to the first obviousness rejection. Moreover, Applicants note that Gao cannot possibly disclose any of the unexpected benefits of the present invention as there is no discussion in Gao of detecting non-whole PTH, as confirmed by Dr. Gao's attached Declaration.

Obviousness Double Patenting Rejection

Claims 23 and 24 have been provisionally rejected under the judicially created doctrine of obviousness type double patenting in view of co-pending Ser. No. 09/344,639.

Response

The present application and the cited one are co-owned. A terminal disclaimer will be filed once claims are allowed. Applicants note the provisional nature of this rejection.

Summary

In summation, Claims 1 to 24 are novel and not obvious, and should be allowed.

If the Examiner has any further questions or reservations regarding this Amendment, Applicants request that the Examiner call their attorney at 843-272-1471.

Respectfully Submitted:

Brian D. Voyce

Attorney for Applicants
Reg. No 28,917
(843-272-1471)

Date: February 7, 2001